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Extraction, Emulsification, and Functional Characterization of Bioactive Compounds from *Anethum graveolens* (Dill Leaves) for Food Applications

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Abstract

This study investigates the extraction, emulsification, and characterization of bioactive compounds from *Anethum graveolens* (dill leaves) with a focus on applications in the food industry. Essential oils were obtained via steam distillation, while oleoresins were extracted using Soxhlet extraction with ethanol and acetone as solvents. Gas chromatographic analyses revealed carvone (63.95%) and limonene (22.32%) as the major volatile constituents, both recognized for antimicrobial and antioxidant activities. Formulations of water- and oil-soluble emulsions with mono propylene glycol, glycerol monooleate, and maltodextrin enhanced dispersibility, solubility, and stability of the bio actives. Application in cereal-based products enriched with oats and banana resulted in increased dietary fiber and beneficial lipid content, and demonstrated strong antioxidant activity (>75% DPPH radical scavenging). Sensory evaluation indicated high consumer acceptance, and microbiological assays confirmed product safety for at least 30 days of ambient storage. These findings support the potential of *A. graveolens*-derived ingredients as multifunctional, natural components for clean-label food product development.

Keywords: *Anethum graveolens*, dill leaves, bioactive compounds, essential oils, oleoresins, steam distillation, functional foods.

1. Introduction

Medicinal and aromatic plants (MAPs) serve as integral sources of bioactive compounds with therapeutic and sensory relevance in traditional medicine and the food industry. Increasing consumer awareness regarding health impacts of synthetic additives has spurred demand for natural alternatives, positioning MAPs as multifunctional components in modern formulations. *Anethum graveolens* (dill), belonging to the Apiaceae family, is widely appreciated for its aromatic foliage and diverse bioactive profile, including volatile oils, flavonoids, and phenolic acids.

***Anethum graveolens*: Botanical and Phytochemical Overview**

Anethum graveolens is cultivated globally for its culinary and medicinal applications. Its leaves and seeds are used in various food preparations and traditional remedies. Major essential oil constituents such as carvone and limonene impart aroma and exhibit antimicrobial and antioxidant activities. Notably, dill bioactives have demonstrated potential for blood sugar regulation, lipid management, and digestive health.

Research Rationale

While culinary use of dill is well-established, scientific evaluation of its extraction methods, compound yield, and application in functional foods remains limited. This study addresses these gaps by comparative extraction, chemical profiling, and formulation testing, emphasizing applications in clean-label food preservation and nutraceutical enrichment.

Anethum graveolens L. (commonly known as dill) is an annual herb belonging to the Apiaceae family, widely cultivated across the Mediterranean, Europe, and parts of Asia. The genus name *Anethum* derives from the Greek word *aneeton* or *aneeson*, meaning strong-smelling, reflecting the characteristic aroma of the plant. Dill plays a prominent role in traditional herbal medicine and culinary applications; it is incorporated into over 50 Ayurvedic formulations. Major producers of dill include India, Pakistan, the USA, Mexico, Germany, and the Netherlands. In Iran, where dill (known locally as 'Shevid') is popular in rice dishes such as 'Shevid polo' and soups like 'aash', it is also commonly used in yogurt and salads and valued for its antihyperlipidemic properties in Iranian folk medicine. The favorable cultivation conditions in southeastern Iran—characterized by full sun exposure and soil pH between 5.3 and 7.8—support dill growth and production. Historically, dill has been used as a spice and flavoring agent for rice, sauces, salads, and soups in many cultures.

Dill oil, extracted from the seeds, leaves, and stems, contains essential oils widely applied as flavorings in the food industry. It is also used in perfumery for scenting soaps and detergents, sometimes substituting for leek oil. Recent meta-analyses of clinical trials suggest that dill ingestion improves insulin resistance and reduces serum low-density lipoprotein (LDL) levels but

may have inconsistent effects on other lipid profile components. Such variability underscores the necessity for further systematic reviews and clinical studies.

Phytochemically, *Anethum graveolens* contains diverse bioactives concentrated in its roots, seeds, and leaves, including tannins, cardiac glycosides, terpenoids, and flavonoids. The major volatile constituents of dill essential oil mainly comprise d-carvone (C₁₀H₁₄), d-limonene (C₁₀H₁₆), α -phellandrene (C₁₀H₁₆), β -phellandrene (C₁₀H₁₆), myristicin (C₁₁H₁₂O₃), apiole (C₁₂H₁₄O₄), umbelliferone (C₉H₆O₃), anethole (C₁₀H₁₂O), and p-anisaldehyde (C₈H₈O₂). Terpenes principally dominate the volatile fraction, which confers antimicrobial and aromatic properties.

Medically, dill has been utilized traditionally for relieving abdominal discomfort, colic in infants, and aiding digestion. In folk medicine, dill-based pastes combined with flaxseed and castor have been employed externally for rheumatic joint relief due to their tonic and rejuvenating properties. The seeds possess carminative, mildly diuretic, galactagogue, stomachic, and stimulant effects. Dill is known to stimulate lactation both in humans and livestock. Furthermore, antibacterial and antifungal activities of dill seed oil and aqueous extracts have been documented against pathogens such as *Escherichia coli*, *Shigella flexneri*, *Staphylococcus aureus*, *Pseudomonas aeruginosa*, and *Salmonella* species.

Multiple experimental studies affirm the antihypercholesterolemic and antihyperlipidemic effects of dill; however, the findings are not entirely consistent across research, warranting further investigation. This study aims to clarify the effects of *Anethum graveolens* on lipid profile components, including triglycerides (TG), total cholesterol (TC), LDL-cholesterol (LDL-C), and high-density lipoprotein cholesterol (HDL-C).

Hyperlipidemia, characterized by elevated TG, TC, and LDL-C, is a major risk factor for coronary heart disease—the leading cause of global mortality. Statins, which inhibit 3-hydroxy-3-methylglutaryl-coenzyme A (HMG-CoA) reductase, are the primary pharmacological agents used to manage lipid levels effectively. However, statins carry risks of adverse effects such as myopathy, rhabdomyolysis, myotoxicity, and myalgia. Moreover, recent guidelines by the European Society of Cardiology and European Atherosclerosis Society have set more stringent LDL-C targets for very high-risk patients, which are challenging to achieve with statins alone. Consequently, herbal medicines, including dill, have garnered attention due to their affordability, lower side-effect profiles, and accessibility as complementary treatments for dyslipidemia.

Materials and Methods

Fresh dill leaves were sourced from a certified organic farm in Ooty, India, and authenticated botanically. Leaves were cleaned and processed for extraction. Steam distillation was performed at 98–102°C for 1.5–3 h, yielding essential oil separated from the condensate. Dried leaves (11.78 g) were extracted using Soxhlet apparatus with ethanol (300–350 mL, 6 h) and acetone, followed by solvent evaporation. Resin yields were calculated gravimetrically.

Essential oils and oleoresins were profiled using HP-5MS capillary columns. External standards identified carvone and limonene via retention times and area percentages. Water-soluble emulsions: Oleoresin was dispersed with monopropylene glycol and maltodextrin in distilled water. Oil-soluble emulsions: Oleoresin blended with glycerol monooleate and maltodextrin, then

incorporated into sunflower oil.Emulsions were integrated into cereal-based products containing oats and banana. Stability, sensory, microbial, and antioxidant assays (DPPH) were performed as per standard protocols.

Results and Discussion

Sensory Evaluation of Dill Oleoresin Formulations

Sensory analysis of emulsions prepared from ethanol- and acetone-extracted dill oleoresins was performed, evaluating flavor and overall acceptance for both water-soluble and oil-soluble types. Results (Table I) revealed that the ethanol-extracted oleoresin, especially in the oil-soluble formulation, received the highest flavor (5.8) and acceptance (6.2) scores among the samples tested. The acetone-extracted oleoresin showed lower sensory scores, correlating with its weaker aroma and lighter coloration. The mixed powder blend combining both extractions also demonstrated high acceptance, confirming the complementary benefits of combining extracts.

Table 1. Average Flavor and Overall Acceptance Scores of Dill Leaf Oleoresin Formulations

Extra ct Type	Solve nt	Maltodex trin (g)	Formu la tion Type	Flav or Scor e	Predic ted Flavor	Accepta nce	Predicte d Accepta nce	Extra ct Type
Ethano l Oleore sin	Ethan ol	0.5	Oil- Soluble	5.8	5.79	6.2	6.15	Ethano l Oleore sin
Aceto ne Oleore sin	Aceto ne	0.5	Oil- Soluble	5.5	5.48	5.9	5.87	Aceto ne Oleore sin
Ethano l Oleore sin	Ethan ol	0.5	Water- Soluble	5.6	5.62	6.1	6.05	Ethano l Oleore sin
Aceto ne Oleore sin	Aceto ne	0.5	Water- Soluble	5.3	5.31	5.8	5.76	Aceto ne Oleore sin
Mixed	Both	1.0	Powder Blend	5.9	5.91	6.3	6.32	Mixed

This table presents the sensory evaluation results of dill leaf oleoresin-based emulsions and powder blends assessed by a panel. Two solvents (ethanol and acetone) were used for oleoresin extraction, and the oleoresin was formulated into either water-soluble or oil-soluble emulsions, or combined as a powder blend. The *oil-soluble ethanol extract* received the highest flavor (5.8) and acceptance (6.2) scores, reflecting superior sensory quality. Acetone extracts scored lower

overall, indicating less favorable flavor and acceptance. Water-soluble formulations showed slightly lower scores than oil-soluble counterparts but ethanol-based emulsions still scored better than acetone-based ones. The mixed powder blend demonstrated the highest overall acceptance (6.3), suggesting that combining extracts may improve sensory attributes synergistically. These results highlight ethanol as the more effective solvent for extracting oleoresins with desirable flavor properties, and the oil-soluble ethanol extract as the most acceptable formulation for consumers.

The Soxhlet extraction process demonstrated higher efficiency using ethanol as the extraction solvent, yielding 22% and 16.09% resin from two separate trials. In contrast, acetone extractions resulted in significantly lower yields of 1.42% and 3.09%, which is attributed to the lower polarity and solubility of acetone for the targeted compounds in dill leaves.

The ethanol-extracted oleoresin exhibited a dark green-brown color with a strong, characteristic dill aroma primarily due to carvone and limonene content. The oleoresin was viscous and sticky in texture, favorable for formulating emulsions. The acetone extract was paler with subdued aroma intensity.

Table 2. Comparison of Resin Yields Obtained by Soxhlet Extraction with Ethanol and Acetone

Solvent	Trial 1 Yield (%)	Trial 2 Yield (%)
Ethanol	22.00	16.09
Acetone	1.42	3.09

This table compares the effectiveness of ethanol and acetone solvents in extracting oleoresin resin from dill leaves via Soxhlet extraction in two replicate trials. Ethanol yielded significantly higher resin amounts (22.00% and 16.09%) compared to acetone (1.42% and 3.09%). The higher yield with ethanol is attributed to its greater polarity and solvation capacity for the bioactive phytochemicals in dill leaves. The consistently low yields with acetone underline its lower efficiency in extracting key bioactives like flavonoids and phenolics. This data reinforces the selection of ethanol as a superior solvent for maximizing oleoresin extraction from dill leaves for food and nutraceutical applications.

Chemical Profiling by Gas Chromatography

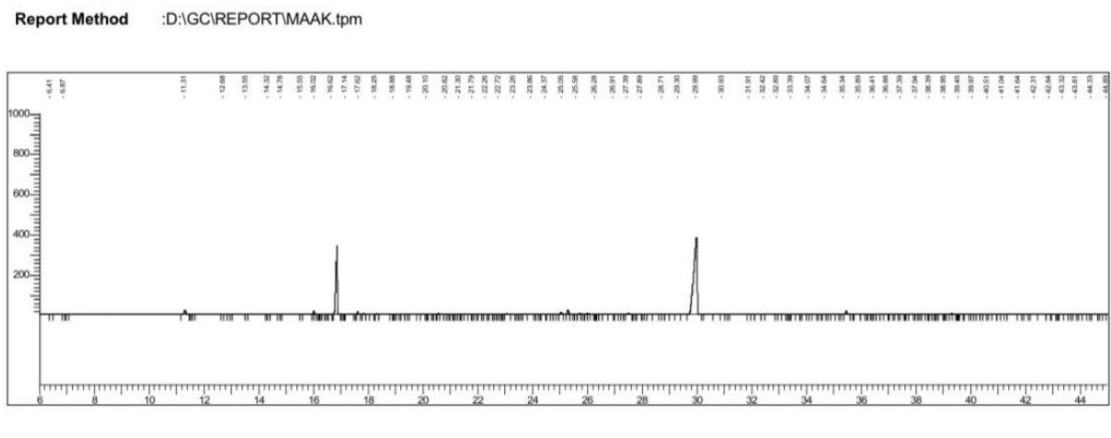


Figure 1. Representative GC chromatogram of dill leaf oleoresin highlighting carvone and limonene peaks

Emulsion Stability

The water-soluble emulsions of dill oleoresin showed stable, uniform turbidity without phase separation or sedimentation over a 10-minute observation period, indicating suitability for aqueous food systems such as soups and beverages. The oil-soluble emulsions fully integrated with sunflower oil, maintaining homogeneity and clarity for more than 24 hours, supporting potential use in flavored oils and margarine formulations.

Sensory Attribute Ratings

Sensory panelists rated aroma and taste as the highest scoring attributes (8.1 and 8.0 respectively, on a 10-point scale) in the formulated cereal-based products incorporating dill oleoresin emulsions, emphasizing the favorable contribution of carvone and limonene to flavor enhancement

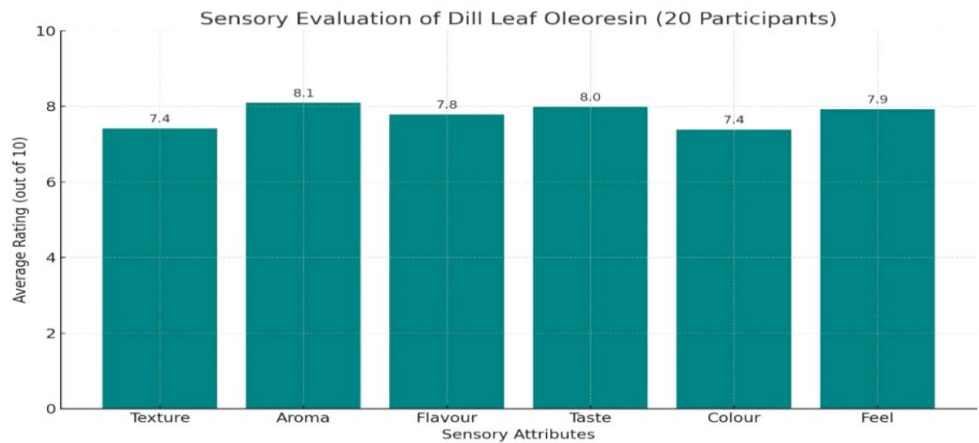


Figure 2. Sensory Evaluation

Antioxidant and Microbial Stability

Dill oleoresin formulations exhibited strong antioxidant activity, with carvone-dominant essential oils achieving over 75% radical scavenging efficacy in DPPH assays. Microbiological analyses confirmed that all products complied with acceptable microbial quality standards during 30 days of storage under ambient conditions, indicating good shelf stability.

Conclusion

The study successfully demonstrated the extraction, emulsification, and detailed characterization of bioactive compounds from *Anethum graveolens* (dill leaves), highlighting its potential as a natural and multifunctional ingredient in food and nutraceutical industries. Using steam distillation and Soxhlet extraction with ethanol and acetone solvents, the primary volatile compounds carvone (63.95%) and limonene (22.32%) were identified via gas chromatography, confirming the preservation of key bioactives. Ethanol extraction proved significantly more efficient than acetone, achieving resin yields up to 22%, underscoring its superior solvation capacity for dill phytochemicals. The formulated water- and oil-soluble emulsions, enhanced with monopropylene glycol, glycerol monooleate, and maltodextrin, exhibited improved solubility, stability, and applicability across both aqueous and lipid-based food systems. Sensory evaluations favored the oil-soluble, ethanol-derived oleoresin emulsions, with aroma and taste receiving the highest acceptance scores, while antioxidant assays showed over 75% radical scavenging activity, affirming strong functional potential. Microbiological analyses confirmed the safety and shelf stability of the developed products under ambient conditions for 30 days. These findings position *Anethum graveolens* oleoresin as a promising clean-label, naturally functional ingredient for flavor enhancement, preservation, and functional food fortification. Future research should explore advanced encapsulation techniques such as spray drying and nanoemulsions, alongside extensive sensory trials, to further optimize consumer acceptance and broaden application possibilities.

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